CLAIMS

1.-8. (Canceled)

9. (Previously presented) Process for the production of a highly active glycoprotein, comprising:

expression of a highly active glycoprotein in an expression cell line, harboring at least one defect in the sugar nucleotide biosynthetic pathway of sialic acids and which is transfected with nucleic acid encoding the glycoprotein, in a medium supplemented with a concentration of at least one sialic acid precursor additive, the concentration being determined by a process comprising:

(i) expression of a plurality of different sialylation forms of said glycoprotein by differential sialylation using different concentrations of at least one sialic acid precursor;

and

(ii) determination of the activity of the different sialylation forms in comparison with a reference glycoprotein in (a) suitable bioassay(s);

and

(iii) selection of the sialylation form with the higher/highest activity and determination of the concentration of the sialic acid precursor additive(s) which is correlated with the higher/highest activity level of said glycoprotein.

10. (Canceled)

11. (Canceled)

12. (Withdrawn) Process for the generation of an expression cell line with a defect in the sugar nucleotide biosynthetic pathway of sialic acids comprising the selection of expression cell line from primary cells or cell lines with a recognition molecule that binds to desialylated structures which can be sialylated by at least two enzymes.

13. (Withdrawn) Process of claim 12, wherein the cells from primary cells or cell lines are mutagenized before selection.

14. (Withdrawn) Process of claim 12, wherein the structures are O-glycans.

15. (Withdrawn) Process according to claim 12, wherein the desialylated structures can be sialylated by alpha2-3 and alpha2-6 bound sialic acids.

16. (Withdrawn) Process according to claim 12, wherein the recognition molecule is a lectin or a carbohydrate specific antibody.

17. (Withdrawn) Process according to claim 12, wherein the recognition molecule is a lectin or a carbohydrate specific antibody recognizing the core-1 structure.

18. (Withdrawn) Process according to claim 12, wherein the expression cell line is derived from the group comprising Per.C6, HEK293, K562, CV1, COS-7, Hybridoma cells, Namalwa, BHK and CHO.

19.-23. (Canceled)

24. (Previously presented) The process of claim 9, wherein a partially sialylated glycoprotein is produced.

25. (Previously presented) The process of claim 9, wherein the defect in the biosynthetic pathway of sialic acids is a loss-of-function of a protein involved in the sugar transportation.

26. (Previously presented) The process of claim 9, wherein the defect in the biosynthetic pathway of sialic acids is a loss-of-function of a protein involved in the sugar transportation, and wherein the protein involved in sugar transportation is selected from the group consisting of CMP-sialic acid transporter, a kinase in the biosynthesis of CMP-sialic acid, a dehydrogenase in the biosynthesis of CMP-sialic acid, a phosphatase in the biosynthesis of CMP-sialic acid, a synthetase in the biosynthesis of CMP-sialic acid, a transketolase in the biosynthesis of CMP-sialic acid, a transaldolase in the biosynthesis of CMP-sialic acid, an isomerase in the biosynthesis of CMP-sialic acid, a transferase in the biosynthesis of CMP-sialic acid, and an epimerase in the biosynthesis of CMP-sialic acid.

27. (Previously presented) The process of claim 9, wherein a sialic acid precursor additive is used which results in glycoproteins with natural sialic acid modifications.

28. (Previously presented) The process of claim 9, wherein the defect in the biosynthetic pathway of sialic acids results in a decreased or absent enzymatic activity of UDP-N-acetylglucosamine-2-epimerase.

29. (Previously presented) The process of claim 9, wherein the glycoprotein is secreted by the cells of the expression cell line.

30. (Previously presented) The process of claim 9, wherein the defect in the biosynthetic pathway of sialic acid is a mutation of an epimerase.

31. (Previously presented) The process of claim 9, wherein the expression cell line is selected from the group consisting of NM-F9 (deposited under the accession number DSM ACC2606), and NM-D4 (deposited under the accession number DSM ACC2605).

32. (Previously presented) The process of claim 9, wherein the glycoprotein is selected from the group consisting of Glycophorin A, EPO, G-CSF, GM-CSF, FSH, hCG, LH, an interferon, an interleukin, an antibody, or one or more fragments of said glycoporteins.

33. (Previously presented) The process of claim 9, wherein at least one sialic acid precursor additive is selected from the group consisting of ManNAc, acetylated ManNAc, peracetylated ManNAc or fetuin.

34. The process of claim 9 wherein GM-CSF is expressed in the expression cell line NM-F9 (deposited under the accession number DSM ACC2606), and the medium is supplemented with the sialic acid precursor additive ManNAc in a concentration of 90 mM.

35. (Previously presented) Glycoprotein GM-CSF producible by the process of claim 34.

36. (Withdrawn) Process for determining a desired concentration of at least one sialic

acid precursor additive for expression of a highly active glycoprotein in an expression cell line, harboring at least one defect in the sugar nucleotide biosynthetic pathway of sialic acids and which is transfected with a nucleic acid encoding the glycoprotein, in a medium supplemented with said desired concentration of at least one sialic acid precursor additive, comprising:

(i) expression of a plurality of different sialylation forms of said glycoprotein by differential sialylation using different concentrations of at least one sialic acid precursor; and

(ii) determination of the activity of the different sialylation forms in comparison with a reference glycoprotein in (a) suitable bioassay(s); and

(iii) selection of the sialylation form with the higher/highest activity and determination of the concentration of the sialic acid precursor additive(s) which is correlated with the higher/highest activity level of said glycoprotein.

37. (New) Process of claim 9 wherein under (iii) a partially sialylated glycoprotein having an activity higher than a reference glycoprotein is selected, and wherein said reference glycoprotein has a higher degree of sialylation than the partially sialylated glycoprotein.